19. Mendelian Experiments on Fowls. III. Production of Dominant Pile Colour.

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[Received March 1, 1922 : Read April 4, 1922.]

(Plates I. & II.)

Two previous papers by me on Mendelian experiments in Fowl breeding have been published in the 'Proceedings of the Zoological Society.' The last, in the volume for 1919, described the production of a recessive pile from a cross between a Silky hen and a *bankiva* cock. The present paper records the results of a cross between a dominant white, namely White Leghorn, and Black-red Game, made with the object of finding whether a dominant pile would result.

In March 1919 I purchased from a Yorkshire breeder a Black-red Game cock, which was stated to be pure-bred, and mated it with two white Leghorn hens kindly supplied by Mr. Seth Smith from poultry in the Society's possession. From these parents two broods of chickens were hatched—the first on April 27–28th, the second about May 10th. There were eight chicks in each brood.

The colour of the down of these chicks was canary yellow, darker round the neck, and diminishing to white behind and below, but none were pure white all over. There was no sign of stripes. Bateson ('Principles of Heredity,' 1909, p. 102) states that the F_1 offspring of White Leghorn by Indian Game or Brown Leghorn is, when newly hatched, *white* with a few specks of black. There may be some breeds of fowls which are pure white in the down, but all that I have studied are not white but yellow or fawn colour. It is well known that young ducklings of white breeds are yellow in the down. Several of the chicks of my cross had one or more black spots and others had black and red specks on the head, but I was not able at the first examination to take them up and examine them closely. The absence of striping is interesting. The chicks in the down of the Japanese Long-tails which I bred some years ago, and, I believe, the chicks of Black-red Game and of Game fowls, generally have a median dorsal dark brown stripe and two lateral stripes of the same colour, the rest of the down being fawn colour or buff. The recessive or non-coloured chicks from the cross of Silky by *G. bankiva* had yellow down on the body generally with white stripes in place of the dark stripes of the coloured chicks. According to Bateson the chicks of pure-bred Silkies have the same marking as this. It would seem, therefore, that in the cross White Leghorn by Black-red the dominance of the white

prevents the dark striping of the chicks, though it does not prevent the occurrence of spots and specks of black or red. On the other hand, Bateson (*loc. cit.* p. 120) states that in the down the chickens of Pile fowls have longitudinal stripes of light chestnut.

On June 4 I made a more thorough examination, but the chickens were wild and difficult to catch, and on reading my notes I found I had only handled 13 out of 16. The first brood were now almost fledged, the second about half-fledged, so that the colour recorded now is that of the first plumage of true feathers. The condition observed was as follows:—

In three no colour was seen; feathers all white.

Four had much brown colour on back, wings, and breast.

One had numerous scattered black feathers.

Five were white with one or more black specks.

The birds were examined thoroughly about six weeks later, namely on July 16 when they were completely fledged, and the sex was recognisable from the size of the combs, although the tails of the cocks were not fully developed.

Cocks, total number 8.

Four of these may be described as imperfect piles; their coloration was:—

Numerous red patches on the saddle and backs of the wings, slight yellow on neck hackles, and very slight on loin hackles; the rest of the plumage white. Legs white with yellowish tinge.

The other four had much less colour, and some of them had more black:—

5. Same marking, but golden yellow instead of red; only a few red feathers on the wings.

6. Black spots on saddle and back of wings, one black spot on the breast, and one on neck. Slight golden yellow on loin hackles.

7. Only a few small red patches on back of wings, but three or four single black feathers; also a tinge of gold on saddle.

8. Black and red ticks on back of wings and back, more black on the right side.

Hens, total number 7.

Four of these had a great deal of reddish-brown colour, different from that of the cocks, which was a brighter red. This colour was "laced" all over back, and saddle hackles, more continuous and uniform on back of wings. There was uniform brown on throat, breast, and abdomen, but pale, *i. e.* diluted with white. Only the end of the tail was quite white. (Pl. I. fig. 2.)

This coloration agrees fairly well with the description by Mr. Douglas of hen piles in Wright's 'Book of Poultry,' 1885:—"Head light golden chestnut; hackle white, laced with yellow chestnut; back a creamy white ground-colour, slightly laced, a shade of gold prevailing; salmon-coloured wing-coverts almost

similar to back, but just a little heavier in the dark colour; breast a rich chestnut right up to the throat, running off to a white, but not pure on the thighs; tail almost a pure white."

5 and 6. No brown but scattered dark specks, many of the feathers in these with minute points of black all over, appearing grey. No. 5 with more black than No. 6 but of same type.

7. Deep black isolated feathers on wings and back, with slight trace of red, uniform over back.

One chick had been lost.

It will be seen, therefore, that on July 16th in these F_1 birds there were two groups—one of four cocks and four hens of the pile type, and the other of four cocks and three hens with small scattered spots of black and red. No. 5 among the cocks belonging to the second group is more similar to the pile, but with colour much less developed.

Subsequently the red colour in the cocks increased considerably, and the pile coloration was more completely developed. Thus on Sept. 27 the birds were nearly full grown and with fully-developed plumage. There were six cocks that looked like fairly typical piles, *i. e.* white birds with rich red backs. In addition there was one with similar marking, but straw colour on the back instead of red, and another with rather smaller comb and wattles and no broad area of colour on back, but separate spots, and a lean and game-like shape. The four pile hens were much the same as before, and the other three hens still had only scattered specks of black and yellow.

In December one of the pile cocks was accidentally killed. The last of the cocks described above, which was more backward in development than the others, had by this time developed the pile coloration like the rest, but with pale yellow colour in the back and saddle, not red.

Thus the F_1 generation in this experiment shows that the pile coloration in the cocks is a heterozygote. It may be said that the white of the Leghorn is dominant to the black in the black-red, but not to the red: hence the heterozygote is white-red instead of black-red. The dominance to black is not, however, quite complete, as black specks or occasional black feathers occur. In the hens the pile coloration takes a different distribution, corresponding to the different distribution of black in the hen of the black-red.

F_2 *Generation.* Year 1920.

In Feb. 1920 I mated the F_1 fowls in two separate pens for breeding. In one pen were put one of the most deeply coloured pile cocks with the four typical pile hens. One of the latter died before breeding. In the other pen were one of the palest pile cocks with three hens which were white with spots and specks of colour. A number of eggs, about 25 from each pen, were obtained,

and hatched in incubators about April 14th. The chicks were examined on April 17th, with the following results :—

A. From the pile parents there were 16 chicks, 13 of which were yellow without spots. The other three were coloured.

(1) Bluish primaries and rump, reddish back, sides and belly yellow.
(2) Smoky black on head and back, and on wing-primaries.
(3) Smoky black on back and head.

B. From the pale pile cock with white hens there were 19 chicks : of these 11 had yellow down apparently without colour, and 8, including one that died after hatching, had some colour. Of these 8, four had one or two black spots and four had more continuous colour, the darkest being the dead one, which was dark smoky black over the head and the whole of the back and wings.

The other three were as follows :—

(1) Slightly greenish on rump.
(2) Slightly bluish rump, head ditto, otherwise yellow.
(3) Back dark reddish combined with blackish colour, slight black spot on head.

If we consider these lots separately, the theoretical expectation, on the supposition that the black-red colour of the original male parent was a single character, would be one-fourth black-red in F_2, the black-red being recessive. Thus in the chicks from Pen A, 16 in number, there should have been 4 coloured : the actual number was 3. Of the chicks from Pen B, 19 in number, one-fourth would be between 4 and 5, or very nearly 5. The actual number was 4 if we include only those which had more or less continuous colour. If we take the two lots together, all the F_1's being heterozygote, the number of coloured recessives was 7 out of 35. It would be expected, however, that black-red recessive chicks in the down would be brown with black stripes, like the black-red dominants in my cross between *bankiva* and Silky. There were none at all like this.

On October 9th I made a careful examination of these birds of the F_2 generation, which were then in mature plumage, and their characters were :—

A. From pile parents.

7 Cocks, all showing the pile coloration, but with different degrees of intensity in the colour on the back, ranging from pale yellow to red.

1. Deeply coloured pile ; legs yellow. A photograph of the skin of this bird is shown in Pl. I. fig. 1.
2. Similar but not so deep a colour ; legs yellow.

3. Pale orange on back and hackles, with a few black specks; legs pink.
4. Pale pile; legs yellow.
5. Pale pile; legs pink.
6 and 7. Very pale pile; both with legs pink.

6 Hens. Two of these were coloured all over; three had the pile coloration, *i.e.* feathers laced with fawn colour on the back, the breast and abdomen uniform pale brown; one was white, with trace of reddish lacing on the neck.

1. Almost entirely black, with white edging to feathers on wings and breast and very slight yellowish lacing. (Pl. II. fig. 2.)
2. Grey all over, with slight orange lacing: legs not noted. (Pl. I. fig. 1.)
3. Strong dark pile coloration of hen type; legs pink.
4. Good hen pile, well laced; legs pink.
5. Pile lacing rather pale; legs pink.
6. White with trace of reddish lacing on neck; legs pink.

B. Offspring of cock with slightly yellow back, and hens white with black specks.

8 Cocks. One of these was a moderately coloured pile; six were pale or very pale piles; and one was not a proper pile, but had spots of orange and black on wings, hackles, and tail. Four of them had yellow legs and four had pink legs.

9 Hens. Two of these were typical piles, with pale back laced with fawn colour and brown breast, one rather more deeply coloured than the other. One was reddish brown all over with black specks. The rest, six in number, were white with scattered specks of colour, and two of them had a yellowish tinge on head and neck-hackles. I give below the notes for each of these individually:—

4. White with minute specks of black.
5. Yellow on head and hackle; back and tail with many black spots.
6. Yellow on head and hackle; black specks on back.
7. White with a few grey marks on feathers.
8. Very slight specks of black here and there; otherwise white.
9. White with few black spots.

Five of these hens had pink legs and four had yellow legs.

I have not paid special attention to the inheritance of the colour of the legs in this experiment, but will merely point out the leg colours noted in the F_2 generation. Yellow leg means yellow skin, and what I have called pink leg means white skin. The yellow skin was a character of the White Leghorn hens of the parents in the cross, the white skin a character of the Black-

Red Game Cock. According to Bateson, white skin is dominant to yellow, and I believe this was the case in the F_1 generation in this experiment. In the F_2 generation, omitting one specimen in which the colour of the legs was not noted, there were 19 pink legs to 11 yellow. The number of recessives was therefore considerably in excess of the expected number, namely 11 yellow to 33 pink, or 1 to 3. This is, of course, in a total of 30 individuals of no special significance.

It is evident from these results that the Pile type of coloration is, like the Andalusian, a heterozygote and not a pure character which breeds true, and that it results from crossing the dominant white of the White Leghorn with the Black-red of the Game. Fanciers have stated this before, but they have not recorded the individual results nor the segregation of the F_2 generation. Thus Mr. John Douglas in Wright's 'Book of Poultry,' 1885, states: "You can also get a very rich Pile by putting a Wheaten hen to a White or Pile cock." The Wheaten hen is one of the types of the hen of the Black-red Game. Mr. Douglas also says that Piles breed true to colour, but that now and then a cross of the Black-red is thrown in to give hardness of feather. My results are not in agreement with this statement. Mr. Fred Smalley, of Silverdale, Lancashire, who bred Pile Game for many years, kindly answering enquiries from me in 1913, wrote that Pile was dominant to Black-red, and that when bred together, Piles worked out to too much washed-out colour, but could never produce pure white. For this reason they were crossed with Black-red once in seven years. He also informed me that a cross of White Leghorn hens with a Brown Leghorn cock, or White Wyandotte hens with Partridge Wyandotte cock, would produce birds of the pile coloration, though they might not be up to standard Pile colour.

In the F_1 generation, however, the heterozygotes were not all alike, but varied considerably in the amount of colour shown—in other words, in the degree of dominance of the white character of the maternal parent. The 8 cocks all ultimately developed the pile marking, six of them having red colour on the back of varying intensity, and two having only yellow instead of red, one having straw-colour and the other pale yellow. Of the 7 hens, four were fairly typical Piles, the other three were white with scattered black or grey feathers.

The two groups of F_1's mated to produce the second generation both produced some piles, as might be expected since the birds of F_1 were all heterozygotes. The birds of F_2 would be expected to produce pure dominants (DD), *i. e.* white, piles (DR) and black-red, *i. e.* recessives (RR) in the proportion of 1 : 2 : 1. The deeply coloured hens in the F_2 generation are the recessives: unfortunately there were no deeply coloured, *i. e.* black-red, cocks. The three coloured hens are all very different from each other, and this is an interesting fact. If we regard the black-red coloration as a single character, a recessive allelomorph to the dominant white, we should naturally expect recessive individuals to be closely alike, whereas

here we have one black, one grey, and one reddish brown. The recessive character therefore appears in different degrees in different individuals, and the fact that the black hen has some white on its feathers shows that the segregation is not complete and the recessive character not perfectly pure.

The pile birds, both cocks and hens, were soon after Oct. 9th separated from the rest, and on Nov. 20th I examined the hens which were not pile, in order to satisfy myself whether there were any pure dominants, *i. e.* white without specks of colour. Of Lot A there were none without some lacing to the feathers, as seen from the observations of Oct. 9th, given above. Of the six in Lot B which were not deeply coloured nor pile, there was not one without some trace of colour. Three of them showed slight lacing of colour on the neck-feathers, and all except one of these had a few black or grey specks.

According to these results, then, there were no pure dominants either in Lot A or Lot B of the F_2 generation. Two hens in Lot A had very pale lacing. In the cocks of this lot there were all degrees of intensity in the red colour of the pile. In Lot B there was one cock which was not a proper pile, but had spots of orange and black on its back, and six hens with either a trace of lacing or some specks of colour. If these six hens in Lot B are to be regarded as pure dominants, we have six out of 17 or nearly one in three instead of one in four. This may seem a good approximation to the theoretical proportion, but it is unlikely that all the dominants should be female. The fact, however, is that none of these birds were without evidence of colour, and therefore it cannot be said that there was complete and clear-cut segregation.

As I was unable to make many further experiments with these birds of the F_2 generation, I decided to mate some of the hens with least colour with another black-red cock to see if they behaved as heterozygotes or as pure dominants. Accordingly two of the F_2 hens above described with least traces of colour were mated in March 1921 with a *bankiva* (black-red) cock which was among the birds at that time alive in the Zoological Gardens. From this mating nine offspring were produced and reared. As there were traces of colour in the female parents, and some of the F_1 hens were also white with black ticks, I thought it possible that these F_2 hens would behave as heterozygotes, and not as pure dominants. I therefore rather expected that half the chicks would develop into piles and half into black-reds. All the nine chicks obtained, however, developed in mature plumage into fairly typical piles. There were five cocks and four hens, only one cock and one hen having somewhat paler colour than the rest.

Although the number of chicks obtained is small, I am of opinion that the F_2 hens behaved genetically as dominant: if they had behaved as heterozygotes, I think there would have been at least one, and probably more than one, black-red among the offspring, the theoretical explanation being, of course, half the

number piles and half black-reds. At the same time it is to be noted that the pile coloration in these birds was more pronounced and more uniform than in the heterozygotes of F_1, whereas if the F_2 mothers had been pure dominants, there is no reason why the offspring of F_2 DD's with the RR black-red should have any more colour than the DR's of F_1.

If we regard the dominant white as one character and the black-red colour as its allelomorph, the evidence of these experiments seems to show both imperfect dominance and incomplete segregation. The pile produced by my cross, and probably the pile of fanciers also, is a heterozygote, like the blue Andalusian, and it should produce in the F_2 generation whites, piles, and black-reds in the proportion 1 : 2 : 1. In F_1 the white is not a complete dominant; it suppresses the black colour almost but not quite completely, but the red colour was developed in varying degrees in the piles, eight cocks and four hens, while the other three hens showed only scattered specks of black and red.

In the F_2 generation both groups A and B were the offspring of heterozygotes, the parents of A being of the pile coloration, those of B showing greater dominance of white. The two groups may be compared thus:—

F_2 Group A.	*F_2 Group B.*
Recessives.	Recessives.
2 ♀, one black and one grey, each with some white feathers.	1 ♀ reddish brown with black specks.
Heterozygotes.	Heterozygotes.
5 ♂ deep pile to pale pile.	3 ♂: one moderate pile with a few black marks, two pale piles with spots of black, or orange and black.
3 ♀ dark pile to rather pale lacing.	4 ♀: two typical piles, two yellow on head and hackle, with many black spots.
? Dominants.	? Dominants.
2 ♂ very pale pile.	5 ♂ very pale piles, two with only a slight tinge of yellow on back of wings, perhaps not more than occurs occasionally in Leghorn cocks.
1 ♀ white with trace of reddish lacing in neck.	4 ♀ white with scattered specks of black or grey.

But while, as this table shows, the recessives can be definitely distinguished from the heterozygotes, it is not possible to separate the pure dominants from the heterozygotes. In the whitest birds there are some traces of colour, and the amount of colour forms a continuous series from the typical pile cock and hen to the birds with least colour. In the recessive hens also there are distinct traces of white. The segregation therefore is imperfect.

In the second cross with *G. bankiva* ♂ the F_2 hens with least colour were used, and, as mentioned above, the offspring were all pile, *i.e.* showed more pronounced coloration to a more uniform degree in the different individuals. It may be suggested that this was due to mating with a black-red of different race, but I think it shows that the F_2 white hens had more colour in their heredity than the original White Leghorn hens. On the other hand, the genetics may be more complicated than merely the meeting of a single pair of allelomorphs, dominant white and black-red. Bateson in 1909 concluded from his own breeding experiments that the black-red colour was due to two complementary factors X and Y, and that the dominant white of White Leghorn was due to a suppressing factor S for which birds of that breed are homozygous. One or both colour factors may be present in the White Leghorn, but even when both are present, colour is suppressed by the factor S.

Some dominant white are of the genetic constitution Xx Yy SS: the factors of the Black-red Game may be assumed to be XXYY. My cross will then be:—

Xx Yy SS × XXYY.

Gametes ..... YXS, YxS, yXS, yxS × XY.

F_1 ..... XYXYS, XYYxS, XYyXS, XYyxS.

All these combinations contain both factors for colour, *i.e.* XY, and all contain only one suppressing factor S. This would help to explain why in F_1 colour is not entirely suppressed, but it does not explain why the black is suppressed more than the red. It would also help to explain why the degree of coloration is different in the F_1 birds, for in one combination both X and Y are double, while in the other three XY together are present only once, with either an additional X, an additional Y, or neither. The birds of constitution XYXYS then may be the typical piles, and the others the palest cocks and white hens of F_1. The difference, however, was not sharply marked.

Again, one or both of the White Leghorn hens in my first cross may have been XxyyS. In this case the cross could be analysed thus:—

XxyySS × XXYY.

Gametes..... XyS, xyS × XY.

F_1 ..... XYXyS, XYxyS.

There would be only two combinations, and each would have the compound XY only once, or in the simplex condition as in the two last combinations in the former case. There would probably then be no typical or deeply coloured piles. It is more probable, therefore, that in my cross the constitution of the hens was XxYySS. I do not know whether a White Leghorn of constitution XYXYSS could occur, and therefore will not consider it here.

We may suppose that the mating of the F_2's may be represented thus:—

Group A..... XYXYS × XYXYS.
Group B..... XYYxS × XYyXS.
Group A, gametes... XYS + XY × XYS + XY.
F_2 ..... XYXYSS + XYXYS + XYXYS + XYXY.

The first should be pure white dominants, the next two typical piles, and the last recessive black-red, the suppressing factor being absent.

Group B, gametes... XYS + xYS + XY + xY × XYS + XyS + XY + Xy.
F_2 ..... XXYYSS, XXYySS, xXYYSS, xXyYSS.
2XXYYS, 2XXYyS, 2XxYYS, 2xXyYS.
XYXY, xYXY, XYXy, xXyY.

Thus there would be four combinations of Xx and Yy, namely

XXYY, XXYy, XxYY, and xXyY;

and these would be combined with SS, S, or absence of S. There would be, therefore, in 16 individuals four coloured recessives of varying composition, four pure dominants, and eight heterozygotes or piles. This agrees fairly well with my results, if we assume that the different combinations of the colour components affect the degree of visible colour.

It is evident, however, that the combinations which are recessive for S ought to show no more white than did the original Black red, whereas in my specimens the three recessive hens show distinct evidence of white: this is more in accordance with the assumption of imperfect segregation.

In the second cross the hens with most white and least colour were mated with another black-red. If we suppose that the white hens had more of the colour factors than the original Whites and were of constitution XYXYSS, then the cross would be

XYXYSS × XYXY.
Gametes..... XYS and XY.
Progeny ..... XYXYS.

Thus all the piles in this case will be homozygous for the combination XY, whereas in the first cross many of the piles were heterozygotes for this combination.

Lastly we may consider the possible genetic combinations in the fancier's process of crossing pile with black-red to improve the colour of the former. The cross might be

XYXYS × XYXY.
Gametes ..... XYS + XY × XY.
Progeny ..... XYXYS + XYXY.

In this case the piles would be as before, and there would be no improvement.

But the pile might be

$$XYxYS \times XYXY.$$

Gametes..... $XYS + xYS + XY + xY \times XY.$

Progeny ... $XYXYS + XYxYS + XYXY + XYxY.$

Therefore half the Piles would be improved by being now homozygous for the combination XY instead of heterozygous. At the same time the effect on the Black-red recessives from the cross agrees with what fanciers say of the inferiority of such birds to the pure-bred Black-red, for half of these recessives are heterozygous for the XY combination.

EXPLANATION OF THE PLATES.

Plate I.

Fig. 1. F_2 ♂, Pile cock from cross of White Leghorn hens with Black-red Game cock. Killed March 1921, hatched April 14th, 1920.

Fig. 2. F_1 ♀, Pile hen, rather deeply coloured: back light brown or fawn colour, "laced" but not quite regularly, some white at base of each feather. Neck hackles white, with orange edges to feathers ("lacing"); abdomen uniform pale brown. Killed Dec. 1920, hatched April 28th, 1919.

Plate II.

Fig. 1. F_2 ♀, coloured recessive. General colour diluted black, *i.e.* grey. Neck hackles laced with pale yellow; feathers of back dark grey, laced with pale fawn; tail dark grey; abdomen like the back, but much paler. Killed March 1921, hatched April 14th, 1920.

Fig. 2. F_2 ♀, coloured recessive. General colour sooty black, with no red. Neck hackles black, with streaks of light brown; back mostly black, with slight lacing of pale brown or drab; breast and abdomen pale grey; some white on wings. Killed March 1921, hatched April 14th, 1920.